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RAPID DETECTION OF DIURETICS IN HUMAN URINE: PROMINENT ANTIHYPERTENSIVES IN CLINICAL APPLICATIONS AND KEY REGULATORS OF EXTRACELLULAR FLUID & KIDNEY DISORDERS

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Objectives: Diuretics are agents commonly used in diseases characterized by excess extracellular fluid, including chronic kidney disease, the nephrotic syndrome, cirrhosis and heart failure. Multiple diuretic classes, including thiazide-type diuretics, loop diuretics and K^+-sparing diuretics, are used to treat patients with these diseases, either individually or as combination therapies[1]. Diuretics are often abused by athletes to excrete water for rapid weight loss and to mask the presence of other banned substances. Because of their abuse by athletes, diuretics have been included on The World Anti-Doping Agency's (WADA) list of prohibited substances. Therefore, a sensitive & selective analytical method was developed & validated for the detection of different diuretic classes like thiazide-type diuretics, loop diuretics and K^+-sparing diuretics etc. using sophisticated LC-MS/MS technique in human urine.

Methods: An analytical method involving liquid-liquid extraction and detection by Liquid Chromatography Mass spectrometric analysis (LC-MS/MS) in MRM mode (multiple reaction monitoring) using polarity switching. Diuretics detection method was developed and validated as per ISO17025 & WADA ISL guidelines for LOD (Limit of detection), specificity, robustness, selectivity, carryover and matrix effect.

Results: An analytical method was developed for the detection of 17 diuretics in a run time of 14 minutes using polarity switching. The validation results were found within the acceptable limits. The developed method is able to achieved limit of detection ranging from nanogram & picogram levels for all the targeted diuretics. The method has been successful applied for the confirmatory analysis of diuretics.

Conclusions: This method could be a useful tool for the detection of these prohibited diuretics in sports doping control and in clinical, nephrological & medical field. This method has facilitated the detection of diuretics at very low levels in human urine. The applicability of the method was also verified by analyzing the real excretion study samples from human.

Figure-1 Diuretics Chromatogram